Statistical Approach to the Design and Analysis of Platelet Pharmacokinetic Studies

Exploring the science of uncertainty

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OBJECTIVES:

1. Describe key design and data analysis principles associated with *in vivo* platelet pharmacokinetic studies in normal human volunteer subjects, including sample size estimations.

2. Propose data analysis and reporting method(s).

3. Propose acceptance criteria / data interpretation method(s).

Summary Recommendations

- Plan and Perform an Equivalency Test (non-inferiority)
- Perform a Paired Design (randomize ^{111}In, ^{51}Cr)
- Construct One sided Confidence Interval of Control and Test Difference
- Construct the Maximum Acceptable Difference from the data
  - Recovery Maximum Diff. = Control – Control * 0.667
  - Survival Maximum Diff. = Control – Control * 0.50
- Reject Null Hypothesis if CI does not overlap Maximum Difference for Recovery AND Survival (I.e., Control=Test)
- Sample Size: - It Depends

Equivalency Test

Objective: “Test” platelets are equivalent to “Control” platelets

Superiority/Inferiority Study

$H_0: \mu_{Test} = \mu_{Control}$

$H_1: \mu_{Test} \neq \mu_{Control}$

α risk, β risk (1-power),
δ difference

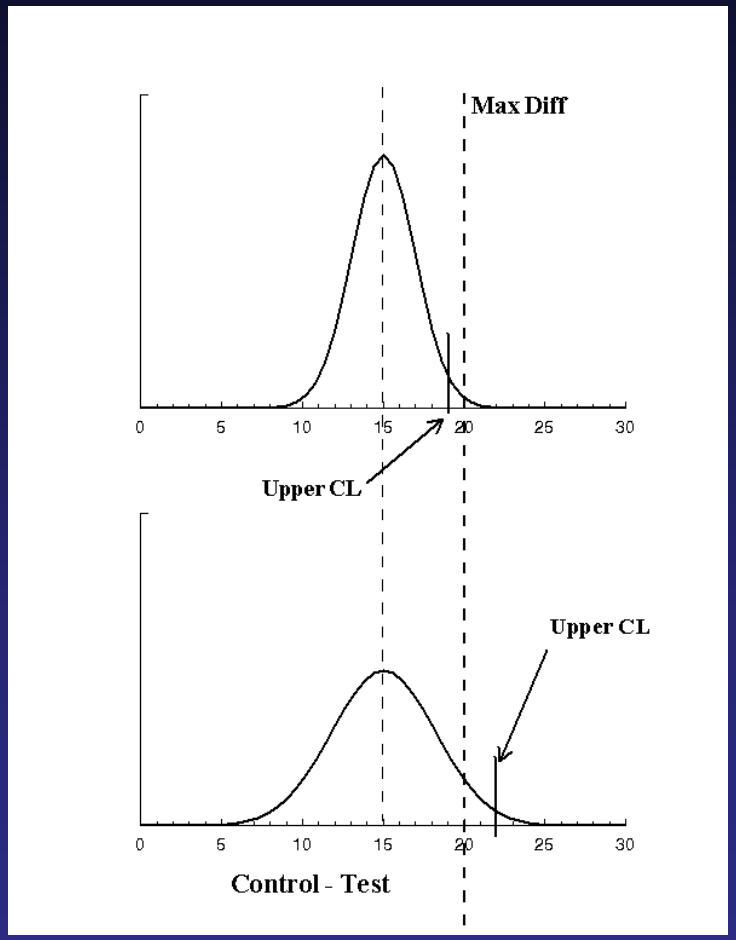
Equivalency Study

$H_0: \mu_{Test} \neq \mu_{Control}$

$H_1: \mu_{Test} = \mu_{Control}$

α risk, β risk (1-power),
δ difference

Equivalency Test – Confidence Interval for the DIFFERENCE



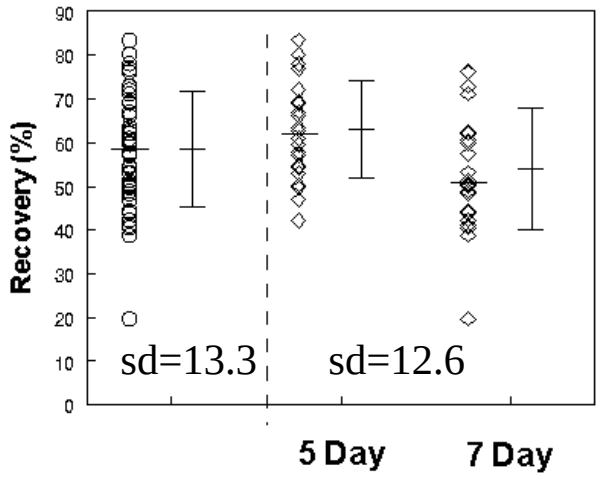
Reject H_0 (accept H_1)

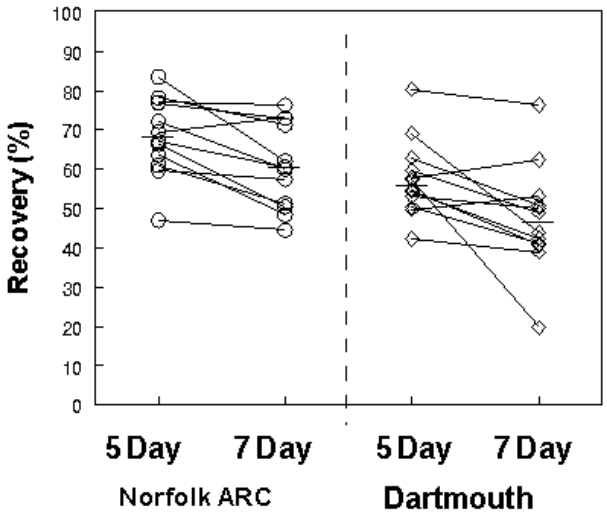
Conclude: Test = Control

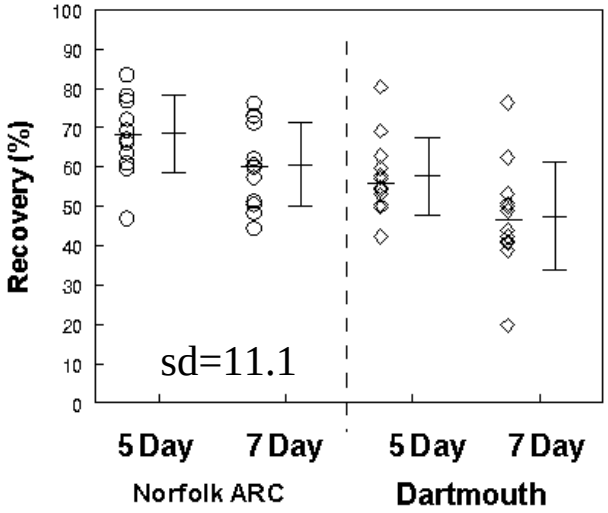
Cannot Reject H_0:

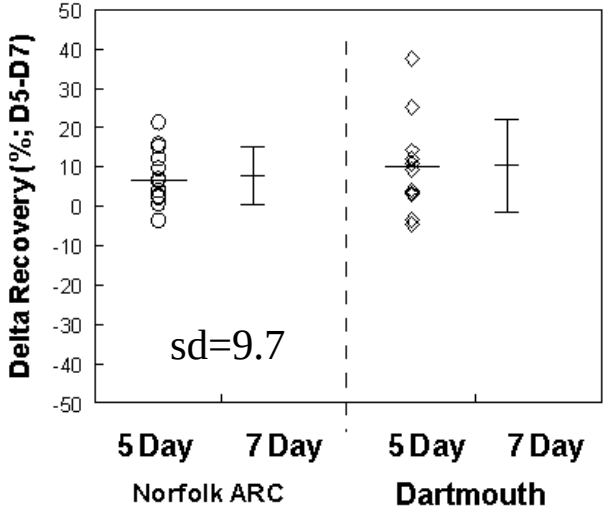
Conclude: There is inadequate evidence that Test = Control

Paired Study Design Reduces Residual Error









Transfusion 2002;42:847-854

Analysis

1. Two Stage Analysis

First Stage

- Adjustments for elution, cell-bound label, baseline (RBC bound)
- Pharmacokinetic model to fit the data (e.g. Multiple-Hit)
- Estimate model parameters (e.g., Recovery and Survival)

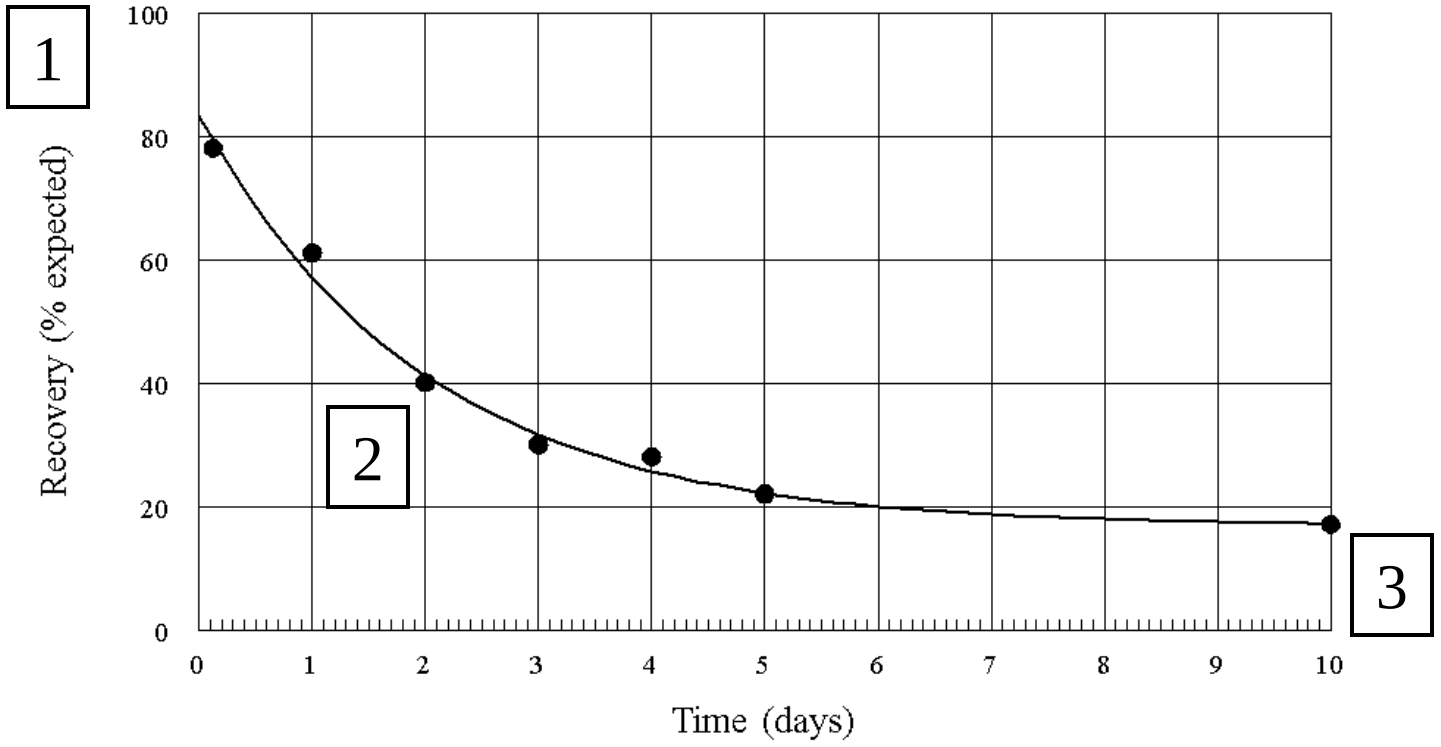
Second Stage

- Analysis of model parameters by paired t-test or regression model

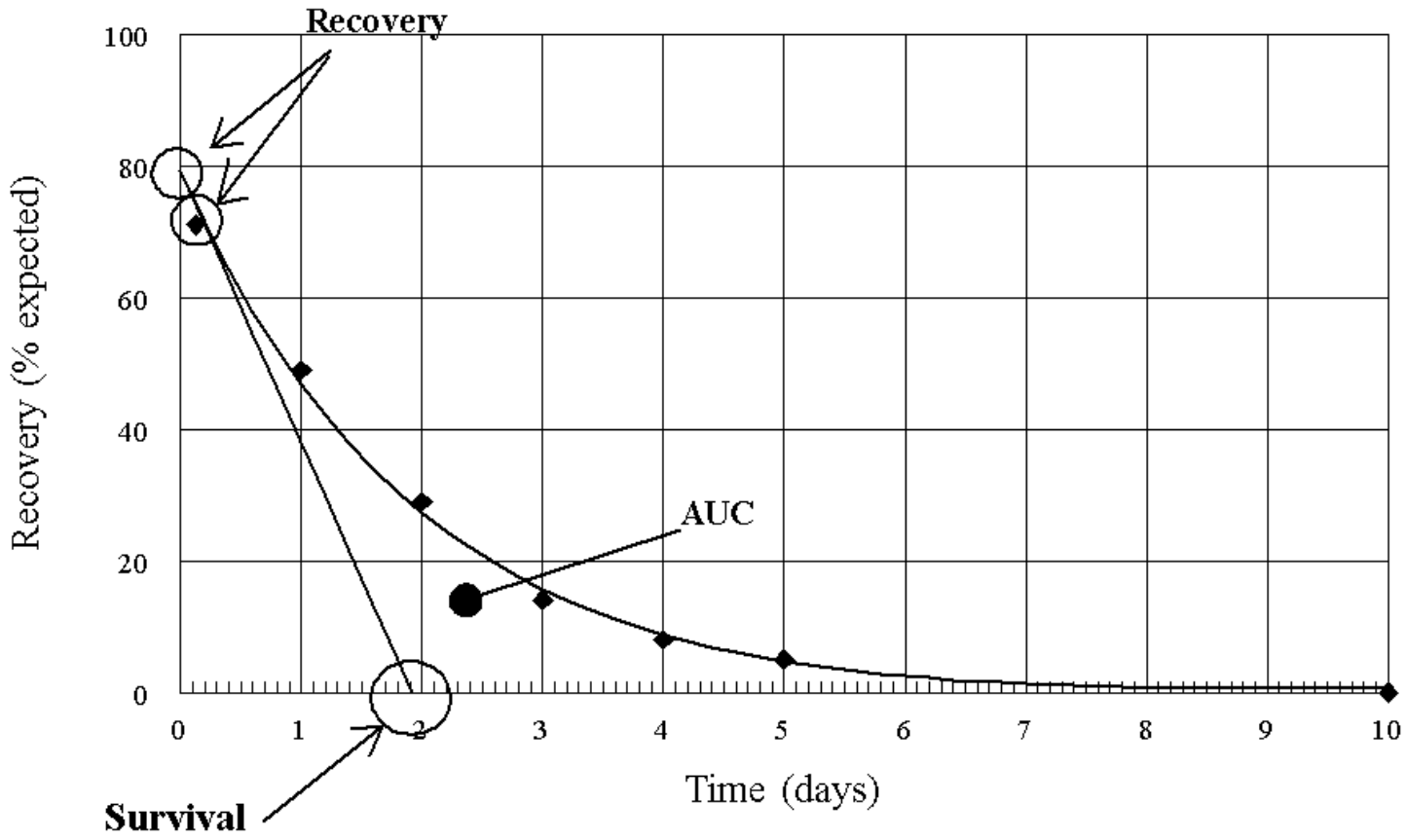
2. One Stage Analysis

- Acceptable and even preferable
- Complex, requires expert
- Two stage simpler and easier to execute

First Stage Analysis: Un-adjusted Data



First Stage Analysis: Fully Adjusted Data



Second Stage Analysis

Confidence interval estimate - Recovery

Subject	Day 5	Day 7	Difference
1	80.2	76.2	4.00
2	68.99	43.94	25.05
.	.	.	.
24	46.89	44.37	2.52
mean=	63.0	53.9	9.0
sd=	11.2	13.8	9.7

Upper Confidence Limit of Difference =

$$\text{Mean} + t_{\alpha,df}\,(sd/\sqrt{n}) = 9.0 + 1.704(9.7/\sqrt{24}) = \mathbf{12.4\%}$$

$\alpha = 0.05$

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Second Stage Analysis

Acceptance Limit - Recovery

Subject	Day 5	Day 7	Difference
1	80.2	76.2	4.00
2	68.99	43.94	25.05
.	.	.	.
24	46.89	44.37	2.52
mean=	63.0	53.9	9.0
sd=	11.2	13.8	9.7

Critical Difference =

Control Mean – Control Mean * 0.667 =

63.0 – 63.0 * 0.667 = **21.0**

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Second Stage Analysis

Hypothesis Test

	Recovery	Survival
Critical Difference	21.0 %	80.4 hr
95% UCL	12.4 %	44.1 hr

12.4 % < 21.0 % AND 44.1 hr < 80.4 hr

Therefore, reject H_0 and accept that Test = Control

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Second Stage Analysis

Statistical package t-test output

Difference	N	Lower 90CL	**Mean**	**Upper 90CL**	Std Dev
Recovery	24	5.63	**9.01**	**12.39**	9.66
Survival	24	10.32	**27.19**	**44.06**	48.22

12.4 % < 21.0 % AND 44.1 hr < 80.4 hr

Therefore, reject H_0 and accept that Test = Control

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Second Stage Analysis

Regression analysis

- More complex and requires one trained in these methods
- Donor should be treated as Random Effect
- Center may be treated as Random or Fixed Effect
- Advantage: regression model may offer opportunity to adjust for other “true” co-variates (e.g., radioisotope, age)

One Stage Analysis

Regression analysis

- Acceptable and even preferable
- Modern, more complex models
  - Non-linear mixed model
  - Donor random effect
  - Center random or fixed effect
- May offer opportunity to adjust for other “true” co-variates
- Requires expert
- Two stage simpler and easier to execute for most

Sample Size – Variance Estimates

		N	Recovery SD	Survival SD
PAIRED				
Holme et al.	BJH 1993;84:717-723			
	Table 2	15	5.49	12.96
	Table 3	16	2.21	13.03
Spectra & Trima	Transfusion. 1999;39:960-6. Transfusion. 2000;40:1214-22.			
		17	4.77	17.46
	Pooled	48	4.37	14.74
Spectra regular & HCP	Transfusion. 2002;42:1333-9.			
		9	9.83	29.65
7 Day Platelet	Transfusion. 2002;42:847-54.			
		24	9.66	48.22
	Pooled	33	9.71	41.12

Sample Size – Variance Estimates

		N	Recovery SD	Survival SD
RATIO (7 Day)				
		24	0.1576	0.3726
UNPAIRED				
7 Day Platelet				
	Fixed Center	24	11.16	42.14
	Random Center	24	29.92	41.52

Estimated Sample Size

$\alpha = 0.05$ $\beta=0.20$ Power=0.80

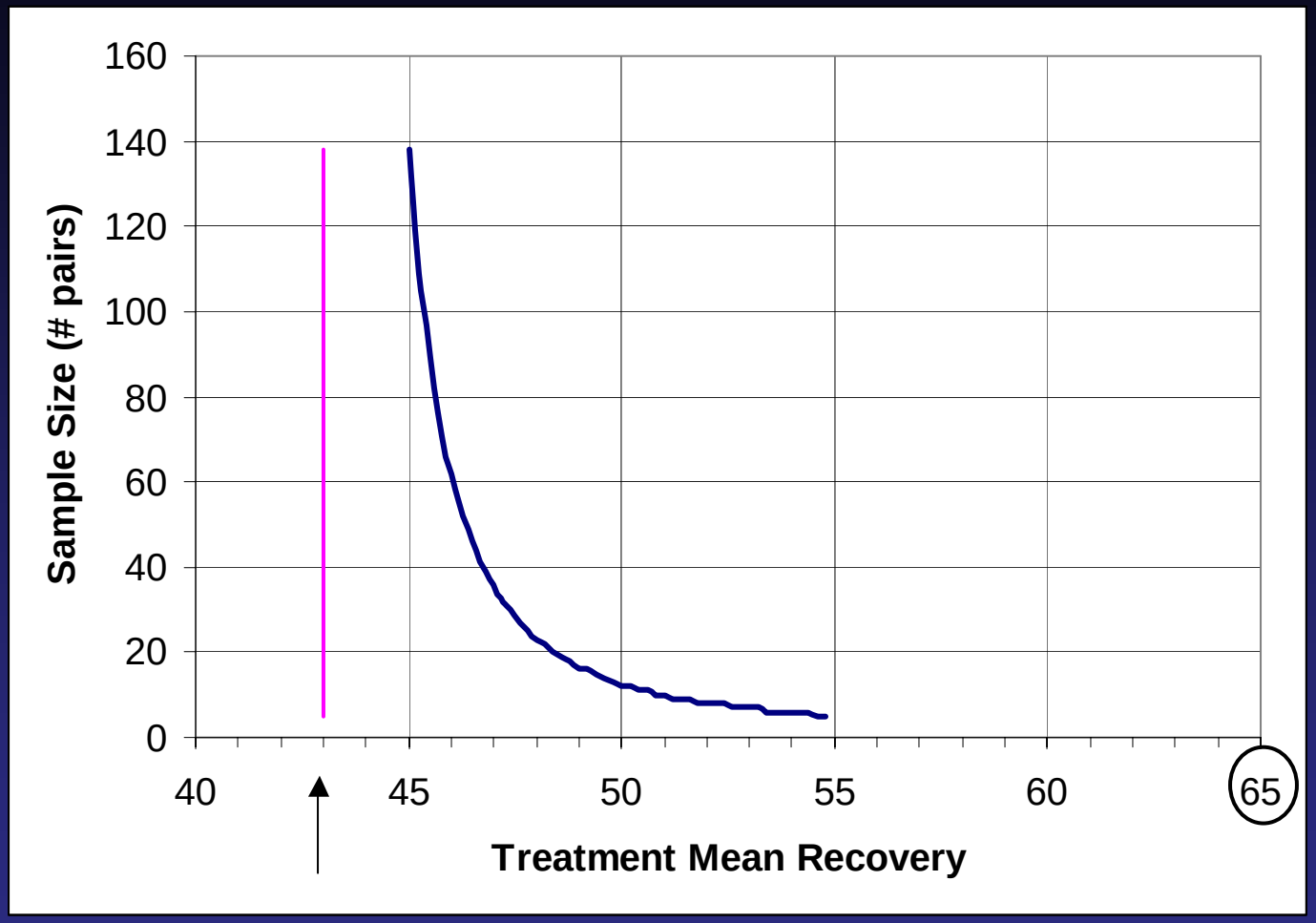
	Recovery		Ratio
Control Mean	**65%**		**1.00**
Treatment Mean	**50%**		**0.77**
Lower Limit	**43% (2/3)**		**0.67**
		SD	**N**
Paired Difference		**9.66**	**13 prs.**
Paired Ratio		**0.16**	**16 prs.**
Unpaired	**Center Fixed**	**11.16**	**33**
	Center Random	**29.92**	**226**

Estimated Sample Size

$\alpha = 0.05$ $\beta = 0.20$ Power=0.80

	Recovery		Survival	
Control Mean	**65%**		**180 hr**	**180 hr**
Treatment Mean	**50%**		**140 hr**	**140 hr**
Lower Limit	**43% (2/3)**		**120 hr (2/3)**	**90 hr (1/2)**
Paired Difference	**SD = 9.66**		**SD = 48.2**	**SD = 48.2**
	N = 13 prs		**N = 37 prs**	**N = 7 prs**

Sample Size Depends on Distance from Minimum Criterion



Additional Recommendations

- Do Not use ratios (Recovery $_{TEST}$ / Recovery $_{CONTROL}$)
  - Increase uncertainty and sample size
  - Model assumptions (e.g., normality) may not hold
- Do Not use one absolute criteria (e.g., Recovery > 43%)
  - Variability in centers, methods, subjects, time is too great
  - Increase sample size

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Regulatory / Scientific Decisions

- **Need Concurrence**
  - Equivalency testing
  - Paired Design with Randomization of labels
  - Non-linear regression model (e.g., COST multiple-hit for 1st stage) – appropriate to describe the data
- **Need an Answer**
  - Simultaneous CI – I.e., recovery and survival must pass?
  - Acceptable difference – Recovery 2/3 Control, Survival ½ Control?
  - Alpha risk – regulators?
  - Beta risk (Power) – up to Sponsor
  - Others: data adjustment, Control, parameters (recovery, survival, AUC?)